(19) European Patent Office

(11) **EP 0 972 522 A1**

(12) **EUROPEAN PATENT APPLICATION**

(43) Publication date:
Jan. 19, 2000 Bulletin 2000/03

(51) Int. Cl.[7]: **A61K 38/34,** A61K 7/48

(21) Application number: **99401720.0**

(22) Application date: **July 8, 1999**

(84) Designated contracting states:
AT BE CH CY DE DK ES FI FR GB GR IE IT LI LU MC NL PT SE
Designated states of extension:
AL LT LV MK RO SI

(30) Priority: **July 15, 1998 FR 9809055**

(71) Applicant: **L'OREAL**
75008 Paris (FR)

(72) Inventors:
- **MAHE, Yann**
91390 Morsang-sur-Orge (FR).
- **Billoni, Nelly**
95750 Valmondois (FR)
- **Breton, Lionel**
78000 Versailles (FR)
- **Bui-Bertrand, Lien**
91600 Savigny-sur-Orge (FR).

(74) Attorney:
TEZIER HERMAN, Béatrice
L'OREAL-DPI
6 Rue Bertrand Sincholle
92585 Clichy Cedex (FR)

(54) **Anti-inflammatory composition**

(57) The invention concerns a composition including at least the association in an effective quantity of a peptide derivative of α-MSH, or of any functional biological equivalent thereof, and of an algae extract of marine origin. The invention also concerns the use of such an association in a composition to combat disorders causing an inflammatory process and a method of cosmetic treatment using such an association.

Description

[0001] The object of the present invention is a composition including, as an active ingredient, at least the association of an effective quantity of a derivative of the melanocyte stimulating hormone of the type α (α-MSH), or of any functional biological equivalent thereof, and of an algae extract of marine origin. The invention also concerns the use of such an association in a composition to combat disorders causing an inflammatory process and a method of cosmetic treatment using such an association.

[0002] Inflammation (or the inflammatory process) is a group of biological reactions found in all levels of animals. In humans, two out of three diseased individuals present an inflammatory syndrome. The inflammation may be localized. It may be defined as the first response to any local injury through a series of nonspecific reactions triggered regardless of the initial cause and occurring in three successive stages: vascular, cellular vascular, and tissue fibrosis.

There is a symptomatic progression of inflammation which ranges from the sensation of skin discomfort, twinges, itching, to swelling, pain, redness, and/or heat. These symptoms are usually due to the infiltration of the injured tissues by edema and/or capillary vasodilatation.

[0003] The signs of inflammation may range to fever, a condition of general malaise, and/or an increase in the concentration of certain blood plasma proteins.

This is a phenomenon which involves, among other things, a series of local cellular reactions and the release of cytokines and other mediators such as substance P, prostaglandins, leukotrienes, bradykinin, histamine, or even serotonin.

Inflammation manifests in a modification of blood flow with, at the site of injury, an increase in vascular permeability resulting in an escape of plasma proteins to the extracellular fluid as well as extravasion of blood cells, primarily of neutrophilic leukocytes and macrophages toward the inflammatory site.

[0004] These phenomena are, in fact, the result of the action of the mediators of inflammation. Among the factors involved in these inflammatory phenomena, it is possible to mention the cytokines, including, in particular, interleukin 1-α, interleukin 1-β, interleukin 6, tumor necrosis factors α and β (TNF-α and -β, the chemokines, such as interleukin 8, or the monocyte

chemotactic activating factor (MCAF), or other chemotactic factors responsible for recruiting lymphocytic, monocytic, Langerhans', or basophilic cells at the level of the inflammatory site, such as leukotrienes B-4, or even other factors involved in the inflammatory cascade, such as arachidonic acid, or the prostaglandins, including especially the E2 prostaglandins.

[0005] Inflammatory phenomena are associated with numerous disorders ranging from simple skin discomfort to pathological conditions.

Examples of skin disorders include sensitive skin, skin discomfort, skin twinges, itching of the skin, swelling of the skin, skin pain, redness, the sensation of heat, erythemas, in particular due to ultraviolet rays, pruritus, erythema nodosum, urticaria, insect bites, allergies, alopecia in the inflammatory phases, joint disorders such as rheumatoid polyarthritis, arthritis, tendinitis, periarthritis, spondyloarthropathies, or the joint disorders of chronic enteropathies, rheumatismal disorders, such as acute articular rheumatism, rheumatoid polyarthritis, pulmonary disorders, such as emphysema, systemic mastocytosis, psoriasis, or even other dermatological disorders such as atrophic polychondritis, erythermalgia, necrobiosis lipoidica. Disseminated lupus erythematosus may also be mentioned.

[0006] Regardless of the phenomena considered, there is a common point to all of these mechanisms which translates into an inflammatory reaction of which the ultimate aspect can be measured by the release by the mastocytic, endothelial, keratinocytic, fibroblastic, melanocytic and/or Langerhans' cells of the skin of at least one inflammation mediator such as histamine, serotonin, heparin, leukotrienes, prostaglandins, cytokines, nitrogen monoxide, or of reactive oxygenated types.

[0007] In particular, it is known that at the level of the surface layers of the skin, in response to a pro-inflammatory signal (chemokines, cytokines, such as interleukin-1), the keratinocytes release interleukin-8 which contributes to the triggering of the inflammatory process.

[0008] For many years, the pharmaceutical industry has sought substances enabling treatment of inflammation. In this regard, many have already been described, known in the literature under the names steroidal anti-inflammatory drugs or nonsteroidal anti-inflammatory drugs (SAIDs or NSAIDs) and whose description is found, for example, in the work by Schorderet and Dayer:

"Pharmacology from the Fundamental Concepts to the Therapeutic Applications", 1992, chapter 37, pp. 541-561, 2nd edition, Frison-Roche/Slatkine Editors.

[0009] Besides the fact that the known anti-inflammatories often present non-negligible adverse effects, it remains interesting to have available new products with anti-inflammatory activity, especially for minor skin disorders, such as sensitive skin, discomfort, twinges, itching, swelling, pain, redness, the sensation of heat, erythemas, in particular due to ultraviolet rays, and pruritus.

[0010] Consequently, the object of the present invention is to be able to have available a new product presenting anti-inflammatory activity and not presenting significant adverse effects.

[0011] This object and others are achieved by the present invention, of which the object is a composition including, as an active ingredient, at least the association of an effective quantity of at least one derivative of the melanocyte stimulating hormone of the type α (α-MSH), or of any functional biological equivalent thereof, and of at least one algae extract of marine origin.

[0012] According to the invention, the peptide derivative is a derivative of the melanocyte stimulating hormone of type α (α-MSH), or melanotropin.

α-MSH was originally described as being produced by the pituitary gland; however, the brain, in general, the blood, the skin, and other tissues are also capable of producing α-MSH.

Thus, in the epidermis, it has been demonstrated by Schauer et al. (J. Clin. Invest. 93, May 1994, pp. 2258-2262) that keratinocytes are a source of α-MSH.

α-MSH receptors are present in numerous cell types, especially in the hair follicles of the human scalp (Pigment Cell Res, 4: 193-8, 1991).

[0013] α-MSH (1-13) is known for its antipyretic activity, its anti-inflammatory activity, and its propigmenting activity. This neuropeptide is known for inhibiting the inflammation induced by cytokines or by other mediators of inflammation as well as by irritants.

The antipyretic signal of α-MSH resides in its carboxy-terminal sequence and can be mimicked by the tripeptide 11-13 carboxy-terminal (L)Lys(L)Pro(L)Val (Watanabe et al. Brain Research Bulletin, Vol. 32, pp. 311-314, 1993).

Thus, the patents US 5,028,592 and WO 88/00833 protect the use of the tripeptide (L or D)Lys-(L)Pro-(L or D)Val in an anti-inflammatory therapeutic treatment method and in the preparation of a medicament to treat inflammation.

[0014] Other derivatives of α-MSH are known for their anti-inflammatory activity. For example, patent application WO 95/08564 describes the anti-inflammatory activity of compounds including at least one sequence of 4 amino acids of α-MSH conjugated with thiocytic acid. That patent application is incorporated herein by reference.

[0015] Thus, in patent application WO 95/08564, it is possible to mention more specifically the following compounds I through VII:

I [(DL) Lip] Glu --- His --- D.homoPhe --- Arg --- Trp --- Gly -- NH2

II [(DH) Lip] Glu --- His --- D.homoPhe --- Arg --- Trp --- Gly -- NH2

III [(DL) Lip] Glu --- His --- ParaFluoroPhe --- Arg --- Trp --- Gly --- NH2

IV [(DH) Lip] His --- D.homoPhe --- Arg --- Trp --- NH2

V [N.Lipoyl-Lysine] Glu ---His --- D.homoPhe--- Arg -- Trp --- Gly -- NH2

VI [N.Lipoyl-Lysine] Hic [sic]--- D.homoPhe Arg Trp Gly NH2

VII [N.Lipoyl-Lysine] His --- D.homoPhe --- Arg --- Trp --- NH2

as well as the derivatives of these molecules in the form of salts of esters or amides.

[0016] The company SEPORGA thus markets a product under the trademark MODULENE® made up of a peptide derivative of α-MSH and having anti-inflammatory properties.

[0017] The applicant has surprisingly and unexpectedly discovered that the anti-inflammatory properties of derivatives of α-MSH can be improved by the association of the latter with an algae extract of marine origin.

[0018] Thus, the applicant was able to demonstrate that the association of a peptide derivative of α-MSH and an algae extract of marine origin presents an anti-inflammatory effect greater than the simple addition of the anti-inflammatory effects which the products taken in isolation can present. Moreover, the applicant has demonstrated that the association produces an anti-inflammatory effect when each of the products of the association is used therein at a concentration for which, used alone, and they produce no effect at all.

[0019] Thus, besides the advantage that the association presents an anti-inflammatory effect greater than that of the products taken in isolation, it enables the use of each of the products of the association at concentrations lower than those used for each of the products taken in isolation.

[0020] In this regard, the following examples illustrate these properties.

[0021] Thus, the first object of the invention is a composition including, as an active ingredient, at least the association of an effective quantity of at least one peptide derivative of α-MSH, or of any functional biological equivalent thereof, and of at least one algae extract of marine origin.

[0022] The term "functional biological equivalent" means a peptide functionally equivalent in terms of biological function of which at least one of the amino acid residues can have been replaced by an amino acid residue with a similar hydropathic index.

The hydropathic index is an index attributed to amino acids as a function of their hydrophobicity and their charge (Kyte et al. (1982), J. Mol. Biol., 157: 105).

[0023] In the area of the amino acids, the geometry of the molecules is such that they can theoretically appear as different optical isomers. There is, in effect, a molecular shape of the amino acid (aa) such that it rotates the plane of polarized light to the right (dextrogyral shape or D-aa), and a molecular shape of the amino acid (aa) such that it rotates the plane of polarized light to the left (levogyral shape or L-aa).

[0024] Nature selected only the levogyral shape for natural amino acids. Consequently, if the peptide used in the compositions according to the invention is of natural origin, it will consist of type L-aa amino acids.

However, chemical synthesis in the lab enables preparation of amino acids with both possible shapes. Starting from this basic material, it is possible, at the time of synthesis of peptides, to incorporate amino acids in both the levogyral or the dextrogyral optical isomer form.

Thus, it is possible, at the time of synthesis of peptides, to incorporate the amino acid residues Lysine-Proline-Valine indifferently in their form D-Lysine (D-Lys), L-Lysine (L-Lys), D-Proline (D-Pro), L-Proline (L-Pro), D-Valine (D-Val), or L-Valine (L-Val).

[0025] Thus, the peptide derivative of the invention can be a peptide whose amino acid residues are indifferently in the form of dextrogyral or levogyral optical isomers.

[0026] Thus, it is possible to mention the peptides containing at least one of the following tripeptides:

D-Lys-D-Pro-D-Val,
D-Lys-D-Pro-L-Val,
D-Lys-L-Pro-D-Val,
L-Lys-D-Pro-D-Val,
D-Lys-L-Pro-L-Val,
L-Lys-D-Pro-L-Val,
L-Lys-L-Pro-D-Val,
L-Lys-L-Pro-L-Val.

[0027] According to the invention, it is, of course, possible to use more than one peptide. In this case, the mixture of peptides may consist of one of the possible combinations of the peptides described above.

[0028] It is possible that, for reasons of resistance to degradation, it may be necessary to use, according to the invention, a protected form of the peptide. The form of protection must obviously be a biologically compatible form. Numerous forms of biologically compatible protections can be envisaged, such as, acylation or acetylation of the amino-terminal end and/or amidation of the carboxy-terminal end.

[0029] Thus, the peptide of the invention may be a peptide in either a protected form or an unprotected form.

[0030] Preferably, according to the invention, protection based on either acylation or acetylation of the amino-terminal end and/or on amidation of the carboxy-terminal end is used.

[0031] In particular, according to the invention, the peptide derivative of the α-MSH is selected from among the peptide derivatives including at least the tripeptide Lys-Pro-Val, the peptide derivatives including at least one sequence of four amino acids of α-MSH conjugated or not with thiocytic acid and, more specifically, the compounds described in the patent application WO 95/08564.

[0032] Preferably, the following compounds I through VII are used:

I [(DL) Lip] Glu --- His --- D.homoPhe --- Arg --- Trp --- Gly -- NH2

II [(DH) Lip] Glu --- His --- D.homoPhe --- Arg --- Trp --- Gly -- NH2

III [(DL) Lip] Glu --- His --- ParaFluoroPhe --- Arg --- Trp --- Gly --- NH2

IV [(DH) Lip] His --- D.homoPhe --- Arg --- Trp --- NH2

V [N.Lipoyl-Lysine] Glu --- His --- D.homoPhe--- Arg -- Trp --- Gly -- NH2

VI [N.Lipoyl-Lysine] His --- D.homoPhe -- Arg -- Trp -- Gly -- NH2

VII [N.Lipoyl-Lysine] His --- D.homoPhe --- Arg --- Trp --- NH2

as well as the derivatives of these molecules in the form of salts of esters or amides.

[0033] A peptide derivative including at least the tripeptide Lys-Pro-Val preferably used according to the invention is the tripeptide Lys-Pro-Val itself, more specifically the tripeptide Lys-Pro-Val for which the Proline amino acid residue is in the non-natural dextrogyral shape (DPro residue).

[0034] Another derivative preferably used according to invention is the derivative sold under the name MODULENE® by the company SEPORGA.

[0035] The peptide used according to invention may, of course, be of natural origin. In this regard, an example which may be cited is α-MSH, widely present in the central nervous system and which can, among other things, be purified from the pituitary gland.

However, with the progress of chemical engineering, it is now easy to synthesize peptides to order, even with great length.

[0036] Thus, the peptide derivative of the invention can be a peptide of either natural or synthetic origin.

[0037] In the composition of the invention, the peptide derivative can be a mixture of peptide derivatives.

[0038] The algae extract of marine origin can be any algae extract of marine origin regardless of the method by which is obtained, provided it meets the criteria defined for the invention, i.e., is

capable of exercising an anti-inflammatory effect on the anti-inflammatory activity of the peptide derivative.

[0039] Preferably, said algae extract of marine origin is an extract of brown algae of the Laminaria family. Even more preferably, the brown algae is an algae of the species *Laminaria digitata*.

[0040] A particularly preferred extract is a solution of oligosaccharides obtained by enzymatic depolymerization of membrane polysaccharides of brown algae, as described in particular in the patent application FR 2753628, incorporated herein by reference.

[0041] In this regard, an algae extract of marine origin particularly preferred according to the invention is an extract sold by the company CODIF INTERNATIONAL, under the name POLYSACCHARIDES ANTI- INFLAMMATION® which is a concentrated solution of an oligosaccharide obtained by controlled enzymatic depolymerization of membrane polysaccharides of a brown algae. It includes the linkage of two uric acids: mannuronic acid and guluronic acid.

[0042] Of course, the composition of the invention is a composition intended for cosmetic or pharmaceutical use.

[0043] The quantity of each of the elements usable according to the invention obviously depends on the effects sought and must be an effective quantity for the association to present the effect sought, in particular an anti-inflammatory effect.

[0044] Thus, to provide one of order of magnitude, the composition of the invention may include the peptide derivative in a quantity by weight representing from 10^{-6} % to 10 % of the total weight of the composition and preferably in a quantity representing from 10^{-3} % to 5 % of the total weight of the composition.

[0045] Likewise, to provide one order of magnitude, the composition of the invention can include the algae extract in a quantity by weight representing from 0.01 % to 10 % of the total weight of the composition and preferably in a quantity representing from 0.02 % to 5 % of the total weight of the composition.

[0046] A further object of the invention also concerns the use, as an active ingredient, of the association in an effective quantity of at least one peptide derivative of α-MSH, or of any

functional biological equivalent thereof, and of at least one algae extract of marine origin in a composition or for the preparation of a composition, with the association or the composition intended to treat inflammation.

[0047] According to this specific aspect of the invention, the association of a peptide derivative of α-MSH, or of any functional biological equivalent thereof, and of an algae extract of marine origin is as previously defined in the text.

[0048] A further object of the invention concerns the use, as an active ingredient, of the association of an effective quantity of at least one peptide derivative of α-MSH, or of any functional biological equivalent thereof, and of at least one algae extract of marine origin in a composition or for the preparation of a composition, with the association or the composition intended to partially or completely inhibit the production of interleukin-8, in particular by the keratinocytes of the skin.

[0049] Examples of disorders causing an inflammatory process were seen previously in the text.

[0050] These inflammatory disorders may be cutaneous or systemic.

[0051] Thus, the compositions using the association of at least one peptide derivative of α-MSH and of at least one algae extract of marine origin according to the invention are intended to combat disorders causing an inflammatory process, and more specifically cutaneous disorders.

[0052] In particular, the compositions according to the invention are intended to combat cutaneous disorders such as sensitive skin, skin discomfort, twinges, itching, swelling, pain, redness, the sensation of skin heat, erythemas, in particular due to ultraviolet rays, pruritus, erythema nodosum, urticaria, insect bites, allergies, alopecia in its inflammatory phases.

[0053] Even more specifically, the compositions according to the invention are intended to combat cutaneous irritations and/or dartres and/or dysesthetic sensations and/or burning sensations and/or pruritus of the skin and/or of the mucosas.

[0054] Regardless of the form of the invention, the composition according to the invention may be ingested, injected, or applied to the skin (on any cutaneous region of the body), the hair, the nails, or the mucosas (buccal, cheek, gingival, genital, conjunctival). Depending on the mode of administration, the composition according to the invention may be presented in all the normally used galenic forms.

[0055] For topical application on the skin, the composition may have the form, in particular, of an aqueous or oily solution or of a dispersion of the lotion or serum type, or of emulsions of liquid or semi-liquid consistency of the milk type, obtained by dispersion of a fatty phase in an aqueous phase (O/W) or vice versa (W/O), or of suspensions or emulsions of a soft consistency of the cream type, or aqueous or anhydrous gel, or even microcapsules or microparticles, or of ionic and/or nonionic vesicle dispersions. These compositions are prepared according to the conventional methods.

They may also be used for the hair in the form of aqueous, alcohol, or hydroalcohol solutions, or in the form of creams, gels, emulsions, mousses, or even in the form of compositions for aerosol, also including a propellant under pressure.

[0056] For injection, the composition may be presented in the form of aqueous or oily lotion or in the form of serum. For the eyes, it can be presented in the form of drops; and for ingestion, it can be presented in the form of capsules, granules, syrups, or tablets.

[0057] The quantities of the different constituents of the compositions according to the invention are those conventionally used in the fields in question.

[0058] These compositions constitute, in particular, cleansing creams, protective creams, treatment and care creams for the face, for the hands, for the feet, for the major anatomical folds or for the body, (for example, day creams, night creams, makeup removing creams, foundation creams, sun screen creams), liquid foundations, makeup removal lotions, body lotions for protection or care, sunburn lotions, lotion gels or mousses for skin care, such as cleansing lotions, sun screen lotions, artificial tanning lotions, compositions for the bath, deodorant compositions including a bactericide, aftershave gels or lotions, depilatory creams, anti-insect bite compositions, pain relieving compositions, compositions to treat certain disorders of the skin such as eczema, rosacea, psoriasis, lichens, severe forms of pruritus.

[0059] The composition according to the invention may also consist of solid preparations constituting soaps or cleansing bars.

[0060] The composition may also be packaged in the form of a composition for aerosol, also including a propellant under pressure.

[0061] The composition according to the invention may also be a composition for hair care, in particular shampoo, waving lotion, treatment lotion, setting cream or gel, a composition of dyes (especially oxidation dyes), possibly in the form of dying shampoos, hair restructuring lotions, a permanent wave composition (particularly in a composition for the first stage of a permanent), and anti-hair loss lotion or gel, and anti-parasitic shampoo, etc.

[0062] The composition may also be for oral/dental use, for example, a toothpaste. In this case, the composition may contain adjuvants and additives standard for compositions for oral use and especially surfactants, thickeners, moisteners, polishing agents such as silica, various active ingredients such as fluorides, in particular sodium fluoride, and possibly sweeteners such as sodium saccharide.

[0063] When the composition is an emulsion, the proportion of the fatty phase may range from 5 % to 80 % by weight, preferably from 5 % to 50 % by weight relative to the total weight of the composition. Oils, waxes, emulsifiers, and co-emulsifiers used in the composition in the form of an emulsion are selected from among those conventionally used in the cosmetic field. The emulsifier and the co-emulsifier are present, in the composition, in a proportion ranging from 0.3 % to 30 % by weight, preferably from 0.5 to 20 % by weight relative to the total weight of the composition. The emulsion may, moreover, contain lipid vesicles.

[0064] When the composition is an oily solution or gel, the fatty phase may represent more than 90 % of the total weight of the composition.

[0065] In known fashion, the cosmetic composition may also contain adjuvants customary in the cosmetic field, such as hydrophilic or lipophilic gellants, hydrophilic or lipophilic additives, preservatives, antioxidants, solvents, perfumes, fillers, filters, odor absorbers, and colorants. The quantities of these various adjuvants are those conventionally used in the cosmetic field, and, for example, from 0.01 % to 10 % of the total weight of the composition. These adjuvants, depending on their nature, may be introduced into the fatty phase, into the aqueous phase and/or into lipid spherules.

As oils or waxes which may be used in the invention, it is possible to mention mineral oils (vaseline oil), vegetable oils (liquid fraction of shea butter, sunflower oil), animal oils (perhydrosqualene), synthetic oils (Purcellin oil), and siliconated oils or waxes (cyclomethicone),

and fluoridated oils (perfluoropolyethers), beeswax, carnauba wax, or paraffin. Is possible to add to these oils fatty alcohols and fatty acids (stearic acid).

[0066] As emulsifiers which can be used in the invention, it is possible to mention, for example, glycerol stearate, polysorbate 60, and the mixture of PEG-6/PEG-32/glycol stearate sold under the name Tefose R 63 by the company Gattefosse.

[0067] As solvents which can be used in invention, it is possible to mention the low alcohols, in particular ethanol and isopropyl, propylene glycol.

[0068] As hydrophilic gellants which can be used in invention, it is possible to mention carboxyvinyl polymers (carbomer), acrylic copolymers such as copolymers of acrylates/alkylacrylates, polyacrylamides, polysaccharides such as hydroxypropyl cellulose, natural gums and clays, and, as lipophilic gellants, it is possible to mention modified clays such as bentones, the metal salts of fatty acids such as the stearates of aluminum and hydrophobic silica, ethylcellulose, polyethylene.

[0069] The composition may contain other hydrophilic active ingredients such as proteins or the hydrolysates of protein, amino acids, polyols, urea, allantoin, sugars and sugar derivatives, hydrosoluble vitamins, plant extracts, and hydroxy acids.

[0070] As lipophilic ingredients, it is possible to use retinol (vitamin A) and its derivatives, tocopherol (vitamin A) and its derivatives, essential fatty acids, ceramids, essential oils, salicylic acid and its derivatives.

[0071] A further object of the present invention is a method of cosmetic treatment, in particular to reduce inflammation, characterized in that a composition such as described above is applied on the skin, on the hair, and/or on the mucosas.

[0072] More specifically, an object of the present invention is a method of cosmetic treatment for a skin soothing effect, characterized in that a composition such as described above is applied on the skin, on the hair, and/or on the mucosas.

[0073] The method of cosmetic treatment of the invention may be implemented, in particular, by applying the hygienic or cosmetic compositions such as defined above, according to the customary technique for use of these compositions. For example: Application of creams, gels, sera, lotions, makeup removing lotions or sun screens or sunburn compositions to the skin or to

dry hair, application of a hair lotion to moisturize hair, shampoos, or even application of toothpaste to the gums.

[0074] The following examples and compositions illustrate the invention without restricting it in any way. In the compositions, the proportions indicated are percentages by weight, unless otherwise noted.

Example 1: Assay of the interleukin-8 induced by interleukin-1 in the surnatant of DK7 cells

Principle and objective of the study:

[0075] This test enables evaluation of the anti-inflammatory potential of various molecules on a keratinocytic cell line (DK7). In this test, an inflammatory situation is mimicked by exacerbating the production of IL-8 of the DK7s by the addition of IL-1α to the culture medium. Then, the effect of a molecule is measured by its inhibiting action relative to this exacerbated production.

[0076] Origin of the cells: Immortalized human cells (infected SV40-T-Ag + Human papillomavirus 16 E6/E7) nontumorous called DK7-Nestlé Research described in patent application PCT/EP 96/05812 (Société des produits Nestlé).

Mode of operation:

[00770] DK7 cells conserved by freezing are first cultured according to conventional protocols in a 75-cm² flask previously coated with a "coating" solution (5 mg of human fibronectin (Sigma) + 5 ml of vitrogen 100 (bovine collagen purified for culture/PC0701/Collagen Corporation) + 50 ml of a 0.1-% BSA solution (BSA/ref.: 343020/Biofluids) + 440 ml of NR1 medium), in the presence of 20 ml of NR2 medium (starting from 500 ml of free serum medium based on NR1 (Biofluids no. P1 85-500), and 2.5 ml bovine pituitary extract (BPE) (Biofluids no. 210) and 5 ml of antibiotic/antimycotic (ref.: 15240-C39/GIBCO)). The cells are then cultured until confluence.

[0078] The cells are then detached from the flask by trypsinization according to conventional techniques. On D = 0, the cells are seeded on precoated 96-well trays at a rate of 200 µl of medium per well (cell density 6×10^4 cells/ml).

[0079] On D = 1, the cells are placed in contact with the product to be tested.

30 to 50 minutes after the treatment, IL-1 at a rate of 2.5ng/ml is added to the culture medium.

The cells are then incubated for 24 hours at 37°C.

The assay of IL-8 in the sumatants as well as an assay of proteins preceded by an XTT/BrdU test are then performed.

IL-8 assay:

[0080] This assay is carried out using an ELISA/IL-8 kit (code RPN 2764/Biotrak/Amersham) according to the supplier's instructions.

The assay is carried out on a volume of 50 µl of culture medium.

The optical density is read at 450 nm using a "Labsystems Multiskan Multisoft" spectrometer.

Protein assay:

[0081] Protein assay is carried out on each sample using a protein assay kit (BCA protein assay kit/ref.: 23225/Pierce) held at ambient temperature.

Products tested:

[0082]

DM1 = MODULENE® at 1 µM;

DM10 = MODULENE® at 10 µM;

Phyco = PHYCOSACCHARIDES ANTI-INFLAMMATION® at 50 µM

DM1 + Phyco = MODULENE® at 1 µM = PHYCOSACCHARIDES ANTI-INFLAMMATION® at 50 µM

DM10 + Phyco = MODULENE® at 10 µM = PHYCOSACCHARIDES ANTI-INFLAMMATION® at 50 µM

Control: Cells treated with IL-1 but not treated with one of the compounds to be tested.

Results:

[0083] The IL-8 assay data are expressed as a percentage of IL-8 per µg of proteins:

	IL-8	% inhibition
Control	44.04	-
DM1 + Phyco	24.73	43.8
DM10 + Phyco	22.71	48.4
DM1	43.69	0.8
DM10	34.89	20.8
Phyco	52.95	0

[0084] The association DM + Phyco significantly inhibits the production of IL-8 in the control batch. The association DM1 + Phyco inhibits the production of IL-8, whereas DM1 alone has no effect on IL-8. This result demonstrates the effect of synergy of the association MODULENE® + PHYCOSACCHARIDES ANTI-INFLAMMATION®.

Example 2: Examples of formulations illustrating the invention. These compositions were obtained by simple mixture of the various components.

[0085]

Composition 1: Day cream:		
	Phycosaccharide anti-inflammation®*	5.00 %
	Modulene®**	1.00 %
	Sucrose stearate	4.00 %
	Stearyl alcohol	2.00 %
	Cyclohexasiloxane	9.00 %
	Mineral oil	4.00 %
	Glycerin	5.00 %
	Xanthan gum	0.30 %
	Carbomer	0.50 %
	Preservatives	0.30 %
	Perfume	0.30 %
	Water	qsp 100
Composition 2: Skin care fluid		
	Phycosaccharide anti-inflammation®*	1.00%
	Modulene®**	1.00%
	Stearyl alcohol	0.40%
	Sorbitan stearate	1.50%
	Glycerin	5.00%
	Xanthan gum	0.20%
	Carbomer	0.10%
	Cyclohexasiloxane	7.00%
	Preservatives	0.30%
	Perfume	0.20%
	Water	qsp 100
Composition 3: Lotion:		
	Phycosaccharide anti-inflammation®*	0.02%
	Modulene®**	1.00%
	Propylene glycol	2.00%
	Cornflower extract	0.10%
	Preservatives	0.10%
	PEG 60 hydrogenated castor oil	0.40%
	Perfume	0.10%
	Water	qsp 100

Phycosaccharide anti-inflammation®*: Oligosaccharide of origin Laminaria digitata (P.M. 3500 Daltons) at 5% in water Modulene®**: lipoyl peptide stabilized by dextran (1 %) in stabilized aqueous solution (Phenonip 0.3 %)

Claims

1. Composition including, as an active ingredient, at least the association in an effective quantity of at least one peptide derivative of α-MSH, or of any functional biological equivalent thereof, and of at least one algae extract of marine origin.

2. Composition according to the preceding claim, characterized in that the peptide derivative of α-MSH is selected from among the peptide derivatives including at least the tripeptide Lys-Pro-Val, with the peptide derivatives including at least one sequence of 4 amino acids of α-MSH conjugated with thiocytic acid.

3. Composition according to the preceding claim, characterized in that the peptide derivative including at least the tripeptide Lys-Pro-Val is the tripeptide Lys-Pro-Val.

4. Composition according to the preceding claim, characterized in that the peptide derivative including at least the tripeptide Lys-Pro-Val is the tripeptide Lys-Pro-Val for which at least the Pro amino acid residue is in the non-natural dextrogyral form.

5. Composition according to claim 2, characterized in that the peptide derivative of α-MSH is selected from among the following compounds I through VII:

I [(DL) Lip] Glu --- His --- D.homoPhe --- Arg --- Trp --- Gly -- NH2
II [(DH) Lip] Glu --- His --- D.homoPhe --- Arg --- Trp --- Gly -- NH2
III [(DL) Lip] Glu --- His --- ParaFluoroPhe --- Arg --- Trp --- Gly --- NH2
IV [(DH) Lip] His --- D.homoPhe --- Arg --- Trp --- NH2
V [N.Lipoyl-Lysine] Glu --- His --- D.homoPhe--- Arg -- Trp --- Gly -- NH2
VI [N.Lipoyl-Lysine] His --- D.homoPhe Arg -- Trp -- Gly -- NH2
VII [N.Lipoyl-Lysine] His --- D.homoPhe --- Arg --- Trp --- NH2

as well as the derivatives of these molecules in the form of salts of esters or amides.

6. Composition according to any one of claims 1 through 5, characterized in that the algae extract of marine origin is an extract of brown algae of the Laminaria family.

7. Composition according to the preceding claim, characterized in that the brown algae is of the species *Laminaria digitata*.

8. Composition according to any one of claims 6 or 7, characterized in that the extract of brown algae is a solution of oligosaccharides obtained by enzymatic depolymerization of membrane polysaccharides of brown algae.

9. Composition according to any one of the preceding claims, characterized in that the peptide derivative is in a quantity by weight representing from 10^{-6} % to 10 % of the total weight of the composition.

10. Composition according to the preceding claim, characterized in that the peptide derivative is in a quantity representing from 10^{-3} % to 5 % of the total weight of the composition.

11. Composition according to any one of the preceding claims, characterized in that the algae extract is in a quantity representing from 0.01 % to 10 % of the total weight of the composition.

12. Composition according to the preceding claim, characterized in that the algae extract is in a quantity representing from 0.02 % to 5 % of the total weight of the composition.

13. Use, as an active ingredient, of the association in an effective quantity of at least one peptide derivative of α-MSH, or of any functional biological equivalent thereof, and of at

least one algae extract of marine origin in a composition or for the preparation of a composition, with the association or the composition intended to treat inflammation.

14. Use, as an active ingredient, of the association in an effective quantity of at least one peptide derivative of α-MSH, or of any functional biological equivalent thereof, and of at least one algae extract of marine origin in a composition or for the preparation of a composition, with the association or the composition intended to partially or completely inhibit the production of interleukin-8, in particular by the keratinocytes of the skin.

15. Use, as an active ingredient, of the association in an effective quantity of at least one peptide derivative of α-MSH, or of any functional biological equivalent thereof, and of at least one algae extract of marine origin in a composition or for the preparation of a composition, with the association or the composition intended to combat skin disorders.

16. Use according to the preceding claim, characterized in that the skin disorders are selected from among: sensitive skin, skin discomfort, skin twinges, skin itches, skin swelling, skin pain, skin redness, the cutaneous sensation of heat, erythemas, in particular due to ultraviolet rays, pruritus, erythema nodosum, urticaria, insect bites, allergies, alopecia in its inflammatory phases.

17. Use according to any one of claims 15 and 16, characterized in that the skin disorders are selected from among: skin irritations and/or dartres and/or dysesthetic sensations and/or heating sensations and/or prurituses of the skin and/or the mucosas.

18. Method of cosmetic treatment, characterized in that a cosmetic composition as described in claims 1 through 12 is applied to the skin, the hair, and/or to the mucosas.

19. Method according to the preceding claim, characterized in that the cosmetic treatment has a soothing cutaneous effect.

European Patent Office	**PARTIAL EUROPEAN SEARCH REPORT** Which, according to Rule 45 of the European Patent Convention, is considered, for the purposes of the subsequent procedure, as the European Search Report	Application no. EP 99 40 1720

DOCUMENTS CONSIDERED RELEVANT

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A	FR 2 733 421 A (OREAL) Oct. 31, 1996 (10/31/1996) *page 2, line 21-page 3, line 3; claim 1* *page 7, line 21-page 8, line 26* ---	1-4, 13, 18	
A,D	US 5,028,592 A (LIPTON JAMES M) July 2, 1991 (07/02/1991) *column 2, line 47-column 3, line 16; claims 1, 2* ---	1-3, 13, 18	
A	FR 2 753 903 A (CODIF INTERNATIONAL SA) April 3, 1998 (04/03/1998) *page 3, line 10-line 34; claim 1* ---	1, 6-8, 13, 18	TECHNICAL FIELDS SEARCHED (Int. Cl. 7) A61K C07K

INCOMPLETE SEARCH

The Search Division feels that the present patent application, or one or more claims, do not comply in with the provisions of the European Patent Convention (CBE) to the degree that a meaningful search on the state-of-the-art cannot be performed. Or only partially with regard to these claims.

Claims which were the object of a complete search:

Claims which were the object of an incomplete search:

Claims which were not the object of any search:

Reason for limitation of the search:

Although claims 18 and 19 concern a treatment method for the human/animal body (Article 52 (4) CBE, the search was conducted and based on the effects claimed for the product / the composition.

Search location:	Date search terminated:	Examiner:
THE HAGUE	September 30, 1999	Charles, D

CATEGORY OF THE DOCUMENTS CITED

X:	particularly relevant by itself
Y:	particularly relevant in combination with another document of the same category
A:	technical background
O:	unwritten disclosure
P:	interleaved document

T:	theory or principle basic to the invention
E:	patent document with a date prior to the filing date, which was not published until that date or a later date
D:	cited in the application
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ANNEX TO THE EUROPEAN SEARCH REPORT
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For any information concerning this Annex, please see Official Journal of the European Patent Office, No. 12/82